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Leaf hydraulic conductance, measured in situ, declines and recovers daily: leaf hydraulics, water potential and stomatal conductance in four temperate and three tropical tree species

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Summary Adequate leaf hydraulic conductance (K_{leaf}) is critical for preventing transpiration-induced desiccation and subsequent stomatal closure that would restrict carbon gain. A few studies have reported midday depression of K_{leaf} (or petiole conductivity) and its subsequent recovery in situ, but the extent to which this phenomenon is universal is not known. The objectives of this study were to measure K_{leaf}, using a rehydration kinetics method, (1) in the laboratory (under controlled conditions) across a range of water potentials to construct vulnerability curves (VC) and (2) over the course of the day in the field along with leaf water potential and stomatal conductance (g_s). Two broadleaf (one evergreen, *Arbutus menziesii* Pursh., and one deciduous, *Quercus garryana* Dougl.) and two coniferous species (*Pinus ponderosa* Dougl. and *Pseudotsuga menziesii* [Mirbel]) were chosen as representative of different plant types. In addition, K_{leaf} in the laboratory and leaf water potential in the field were measured for three tropical evergreen species (*Protium panamense* (Rose), *Tachigalia versicolor* Standley and L.O. Williams and *Vochysia ferruginea* Mart) to predict their daily changes in field K_{leaf} in situ. It was hypothesized that in the field, leaves would close their stomata at water potential thresholds at which K_{leaf} begins to decline sharply in laboratory-generated VC, thus preventing substantial losses of K_{leaf}. The temperate species showed a 15–66% decline in K_{leaf} by midday, before stomatal closure. Although there were substantial midday declines in K_{leaf}, recovery was nearly complete by late afternoon. Stomatal conductance began to decrease in *Pseudotsuga*, *Pinus* and *Quercus* once K_{leaf} began to decline; however, there was no detectable reduction in g_s in *Arbutus*. Predicted K_{leaf} in the tropical species, based on laboratory-generated VC, decreased by 74% of maximum K_{leaf} in *Tachigalia*, but only 22–32% in *Vochysia* and *Protium*. The results presented here, from the previous work of the authors and from other published studies, were consistent with two different strategies regarding daily maintenance of K_{leaf}: (1) substantial loss and subsequent recovery or (2) a more conservative strategy of loss avoidance.

Keywords: cavitation, embolism, photosynthesis, transpiration, xylem.

Introduction

The water pathway from the plant stem to the sites of evaporation in the leaf is critically important for maintaining leaf water balance and allowing stomata to stay open, resulting in carbon capture. However, water transport in the leaf is vulnerable to desiccation-induced changes, including conduit embolism and collapse (e.g., Bucci et al. 2003, Nardini et al. 2003, Cochard et al. 2004, Woodruff et al. 2007, Johnson et al. 2009). Recent research has suggested that these changes may be partially due to variation at the molecular scale, including aquaporin gene expression and protein conformation (Cochard et al. 2007, Kaldenhoff et al. 2008), that result in reductions in leaf hydraulic conductance (K_{leaf}). There has been increasing interest in leaf hydraulics over the past 5 years, and it has been determined that K_{leaf} generally declines with increasing water stress, but that the extent of the decline and the water potentials corresponding to the decline, vary from species to species, even within a particular habitat (Salleo et al. 2001, Brodribb and Holbrook 2003, Hao et al. 2008). Our understanding of the mechanisms responsible for the desiccation-induced decline in K_{leaf} is still far from complete, and is complicated by the interacting effects of light, temperature and water status on K_{leaf}. Voicu et al. (2008), Scoffoni et al. (2008) and Sellin et al. (2008) have all found increases in K_{leaf} with increasing light, and Sack et al. (2004) observed increases in K_{leaf} that were greater than would be expected due only to the reduced viscosity of water with increasing temperature.

Even less well understood is the phenomenon of K_{leaf} recovery while still under negative pressures (Clearwater and Goldstein 2005, Nardini et al. 2008).

The vast majority of studies on the effects of dehydration on leaf hydraulics have been conducted with excised leaves in the laboratory (e.g., Brodribb and Holbrook 2003, Lo Gullo et al. 2003, Nardini et al. 2003, Trifilò et al. 2003, Cochard et al. 2004) and only a few studies have measured leaf hydraulic conductance in situ (Brodribb and Holbrook 2003, 2004, Sack et al. 2003, Aranda et al. 2005, Sellin and Kupper 2007). Even fewer studies have focused on diurnal patterns of leaf hydraulics in the field. Brodribb and Holbrook (2004) found that in *Simarouba glauca* DC. K_{leaf} decreased by about half by midday compared to predawn values and recovered completely by the end of the day. They also found a close correspondence between K_{leaf} vulnerability curves (VC) generated in the laboratory and those generated from measurements in the field. Similarly, Bucci et al. (2003) observed daily decreases and recovery in petiole conductivity in two tropical tree species and also found that at a given value of leaf water potential, laboratory generated VC yielded values of petiole conductivity similar to those measured in field-grown plants.

The objectives of this study were to determine the extent to which different species lose and regain K_{leaf} over the course of a typical summer day and to determine if contrasting species show similar relationships between daily time courses of K_{leaf} and stomatal conductance. The authors also wanted to assess the robustness of the rehydration kinetics method for quickly determining K_{leaf} in the field. Finally, leaf hydraulic VC and leaf water potentials measured in the field were used to predict the daily course of K_{leaf} for other species in which K_{leaf} had not been directly measured in the field.

Materials and methods

Field sites and species

Two field sites were used: one near Corvallis, OR and the other in the Republic of Panama. The first field site was chosen based on proximity to the laboratory, the species that were available at the site, and because a previous study with baseline physiological data for the species studied here was performed over the summers of 2002 and 2003 (Davis 2005). The site was located in McDonald-Dunn Research Forest (44°38′ N and 123°18′ W) and all plants were located in a young forest (clear cut about 15 years prior, Davis 2005) and were within 300 m of each other. The Panamanian study site was in an old-growth forest in the Parque Nacional San Lorenzo on the Caribbean side of the Isthmus of Panama (9°17′ N and 79°58′ W). All measurements were carried out between August and September 2008 at the Oregon site, and between December and February 2003 and 2004 at the Panamanian site.

To represent different plant functional groups, one evergreen broadleaf (*Arbutus menziesii* Pursh.) and one deciduous broadleaf (*Quercus garryana* Dougl.) species were selected at the Oregon field site. Two coniferous species were also selected from the same site; one from the *Pinus* clade (long, needle-like leaves, with no plicate mesophyll; *Pinus ponderosa* Dougl.) and one from the *Larix-Pseudotsuga* clade (shorter, flat needles with differentiated palisade and spongy mesophyll; *Pseudotsuga menziesii* [Mirbel] Franco var. *menziesii*) (Esau 1977, Gernandt et al. 2008). In addition, three evergreen species from the tropical site were selected: *Protium panamense* (Rose) I.M. Johnst., *Tachigalia versicolor* Standley and L.O. Williams, and *Vochysia ferruginea* Mart.

Leaf hydraulic conductance and vulnerability

Leaf hydraulic conductance (mmol m^{-2} s^{-1} MPa^{-1}) was determined using a timed rehydration method described in Brodribb and Holbrook (2003), which is based on an analogy between rehydrating a leaf and recharging a capacitor:

$$K_{leaf} = C \ln(\Psi_o/\Psi_f)/t,$$

where C is the capacitance, Ψ_o is the leaf water potential before partial rehydration, Ψ_f is the leaf water potential after partial rehydration and t is the duration of rehydration. Branches about 30–50 cm long were collected from trees early in the morning before significant transpirational water loss and were transported to the laboratory, recut under water and allowed to rehydrate for at least 4 h. Measurements of leaf water potential (Ψ_L) were conducted over the next 3 days on excised leaves/fascicles for initial values (Ψ_o), and for final values after a period of rehydration of t seconds (Ψ_f). Distilled water was used for rehydration of K_{leaf} samples, and water temperature was maintained between 21 and 23 °C.

For measurement of K_{leaf} in the field, branches (about 10–20 cm in length) were collected from trees, and leaves were then excised for determination of Ψ_o, with no equilibration time (Ψ for leaves on the same shoot typically varied by > 0.1 MPa). Leaf samples from the same branch were then rehydrated for a period of t seconds and Ψ_f was measured. Distilled water was used for rehydration of K_{leaf} samples and all measurements took place in the shade. These measurements (both field and laboratory) were performed on individual leaves of *Quercus*, *Arbutus*, *Tachigalia*, *Protium* and *Vochysia*, small shoots (~ 3 cm long) of *Pseudotsuga* and fasicles (three needles each) of *Pinus*.

Field measurements of K_{leaf} along with corresponding measurements of Ψ_L and stomatal conductance were performed over 4 days in August and September 2008 (*Quercus* and *Pseudotsuga* on August 28th and September 3rd and *Arbutus* and *Pinus* on September 9th and September 16th).

All measurements were made on four to eight leaves from six preselected individual leaves about every 90 min from 530 to 600 h (predawn) until 1600–1630 h PDT. All individual leaves were in open areas, and fully sunlit branches/leaves were chosen for measurement (with the exception of predawn measurements). Both laboratory and field measurements of K_{leaf} were normalized by dividing each K_{leaf} value by the corresponding maximum K_{leaf} for each species and condition (field or laboratory; maximum K_{leaf} = mean of 10 highest values) and reported as relative K_{leaf}. This procedure resulted in several (~ 4–5) individual values of K_{leaf} being greater than one for each species and site (see Figure 1).

Values of C were estimated from pressure–volume curves (Scholander et al. 1965, Tyree and Hammel 1972) using the methods described by Brodribb and Holbrook (2003). Briefly, the Ψ_L corresponding to turgor loss was estimated as the inflection point of the graph of Ψ_L versus relative water content (RWC). The slope of the curve before and following turgor loss provided C in terms of RWC (C_{rwc}) for pre- and post-turgor loss, respectively. Five to six leaves of each species were used to construct pressure–volume curves and to estimate C.

Pressure–volume curves were conducted on individual leaves of the broadleaf species, small shoots (about 3 cm) of *Pseudotsuga* and on fascicles of three needles of *P. ponderosa*. Branch samples of about 30–50 cm, from the same individual leaves that were used for rehydration and measurement of K_{leaf}, were excised early in the morning and recut under water in the laboratory. Branches were allowed to rehydrate for at least 4 h before pressure–volume analyses were performed. Pressure–volume curves were created by plotting the inverse of Ψ_L against RWC with alternate determinations of fresh mass and Ψ_L repeated during slow dehydration of the twig on the laboratory bench until values of Ψ_L exceeded the measuring range of the pressure chamber (−4.0 MPa). Leaf water potential was measured using a pressure chamber (PMS Instrument Company, Corvallis, OR). For normalizing C on a leaf area basis, leaf areas for the broadleaf species and *Pseudotsuga* were obtained with a scanner and ImageJ Version 1.27 image analysis software (Abramoff et al. 2004, National Institute of Mental Health, Bethesda, MD) and needle areas for *P. ponderosa* were determined by multiplying mean needle lengths and circumferences (n = 6 needles per species).

Leaf water potentials and stomatal conductance

Stomatal conductance (g_s) was measured with a steady-state porometer (LI-1600, Li-Cor, Lincoln, NE) and leaf temperatures were measured concomitantly with a fine-wire thermocouple (located in the LI-1600 chamber). One-sided leaf areas of foliage from the porometer measurements were obtained with a scanner and ImageJ. Leaf water potential was measured using a pressure chamber on individual leaves of *Quercus*, *Arbutus*, *Tachigalia*, *Protium* and *Vochysia*, small shoots (~ 3 cm) of *Pseudotsuga* and fasicles (three needles each) of *Pinus*. Measurements of g_s and Ψ_L were conducted on the same dates and over the same time intervals as K_{leaf} measurements and were performed on five leaves of each species (per time interval).

Sampling and statistics

For each species, K_{leaf} data were grouped (binned) over water potential ranges of about 0.3 MPa (i.e., −0.61 to −0.90 MPa, −0.91 to −1.20 MPa, etc.) with the exception of the first bin for each species, which corresponded to 0 to −0.6 MPa (Figures 1 and 2). Each bin contained between two and 14 measurements and a total of 46–56

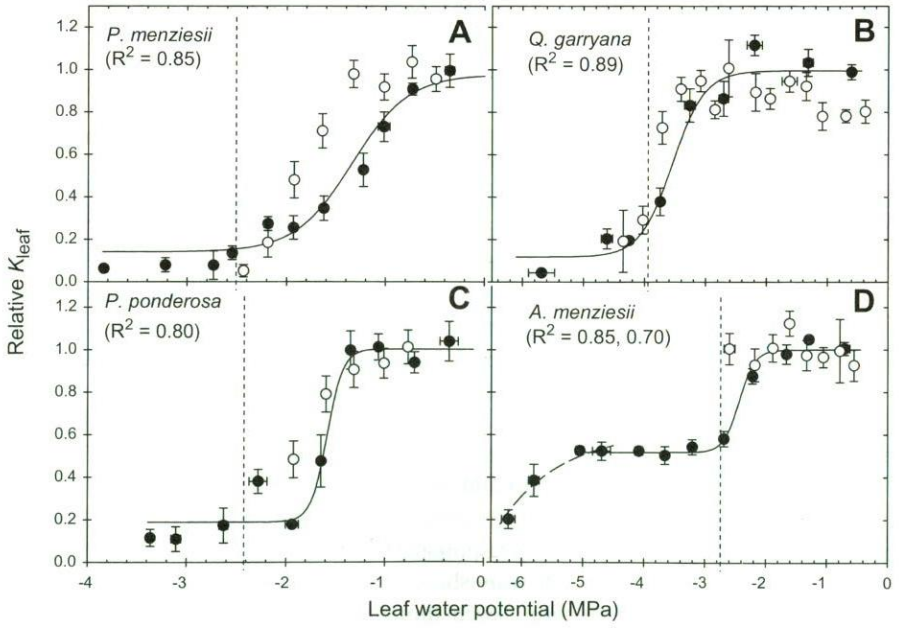

Figure 1. Leaf hydraulic conductance (K_{leaf}) measured in the laboratory (closed circles) and in the field (open circles) for (A) *P. menziesii*, (B) *Q. garryana*, (C) *P. ponderosa* and (D) *A. menziesii*. Dashed lines represent mean bulk leaf turgor loss point (TLP) and vertical and horizontal error bars represent standard error.

Table 1. Species measured, percent of K_{max} at daily Ψ_{min} and the method used to determine percent K_{max} (either measured directly, calculated by measuring transpiration and the difference in Ψ_L in transpiring and non-transpiring leaves, or interpreted from VC/acoustic emission curves (AEC) and Ψ_{min}).

Species	Reference	% K_{max}	Ψ_{min}	%K_{max} method
Prunus mahaleb L.	Kikuta et al. (1997)	13	−2.25	Interp from Ψ_{min} & AEC
Phillyrea angustifolia L.	Kikuta et al. (1997)	72	−1.88	Interp from Ψ_{min} & AEC
Ilex aquifolium L.	Kikuta et al. (1997)	45	−1.46	Interp from Ψ_{min} & AEC
Lauris nobilis L.	Kikuta et al. (1997)	15	−2.65	Interp from Ψ_{min} & AEC
Quercus ilex L.	Kikuta et al. (1997)	64	−1.75	Interp from Ψ_{min} & AEC
Acer campestre L.	Kikuta et al. (1997)	50	−1.61	Interp from Ψ_{min} & AEC
Viburnum tinus L.	Kikuta et al. (1997)	82	−1.29	Interp from Ψ_{min} & AEC
Ceratonia siliqua L.	Kikuta et al. (1997)	90	−1.80	Interp from Ψ_{min} & AEC
C. alliodora	Meinzer et al. (2004)	68	−2.45	Calc by $E/\Delta\Psi_L$
S. morototoni	Meinzer et al. (2004)	64	−1.70	Calc by $E/\Delta\Psi_L$
S. glauca	Brodribb and Holbrook (2004)	30	−1.55	Measured in situ
Byrsonima crassifolia HBK.	Brodribb and Holbrook (2006)	100	−1.3	Interp from Ψ_{min} & VC
Rehdera trinervis Moldenke	Brodribb and Holbrook (2006)	76	−1.2	Interp from Ψ_{min} & VC
Tectaria confluens (F.v.Muell. ex Bak.) Pic.Serm.	Brodribb and Holbrook (2006)	59	−1.15	Interp from Ψ_{min} & VC
Genipa americana L.	Brodribb and Holbrook (2006)	65	−0.85	Interp from Ψ_{min} & VC
C. brasiliense	Bucci et al. (2003)	35	−1.35	Measured (in petioles)
S. macrocarpa	Bucci et al. (2003)	37	−1.30	Measured (in petioles)
Cercis siliquastrum L.	Nardini et al. (2003)	86	−1.85	Interp from Ψ_{min} & VC
Hymeneae stignocarpa Mart. Ex Hayne	Hao et al. (2008)	30	−2.13	Interp from Ψ_{min} & VC
Aegiphila lhotzkiana Cham.	Hao et al. (2008)	34	−1.04	Interp from Ψ_{min} & VC
Myrsine guianensis (Aubl.) Kuntze	Hao et al. (2008)	24	−1.54	Interp from Ψ_{min} & VC
Styrax ferrugineus Nees & Mart.	Hao et al. (2008)	29	−1.41	Interp from Ψ_{min} & VC
Tapirira guianensis Aubl.	FCM, unpublished	9	−1.98	Interp from Ψ_{min} & VC
V. ferruginea	Current study	78	−1.42	Interp from Ψ_{min} & VC
P. panamense	Current study	68	−1.67	Interp from Ψ_{min} & VC
T. versicolor	Current study	27	−2.02	Interp from Ψ_{min} & VC
P. ponderosa	Current study	33	−1.86	Measured in situ
P. menziesii	Current study	31	−2.00	Measured in situ
Q. garryana	Current study	77	−3.64	Measured in situ
A. menziesii	Current study	82	−2.15	Measured in situ
Alnus rubra Bong.	DMJ, unpublished	70	−2.06	Measured in situ

leaves were used for each curve, depending on the species. However, curves were fitted through non-binned data, which reduced the correlation coefficient, but reflected the truer fit of curves through the data. To reflect daily trends in field measurements, data for K_{leaf}, Ψ_L and g_s were grouped according to time across 60 min intervals (Figures 3, 4 and 5). Best fit nonlinear (sigmoid; $y = y_0 + [a/1 + e^{(x-x_0/b)}]$) curves were fit through laboratory K_{leaf} data, R^2 values were calculated and Student's t tests were used to compare the mean of laboratory versus field K_{leaf}.

Results

Overall, K_{leaf} declined with decreasing Ψ_L in all species (Figures 1 and 2). Losses in K_{leaf} with decreasing water potential were similar in laboratory and field measurements, although there was greater variation in K_{leaf} at a given Ψ_L in the field (Figure 1). Maximum K_{leaf} was greater in the field than in the laboratory for all four species in which it was measured under both conditions (Table 2) with a mean increase of 47% from laboratory to field. *Pseudotsuga* displayed the greatest difference between laboratory and field maximum K_{leaf} (88%), while *Arbutus* had the smallest difference (3%).

In the field, measured K_{leaf} increased from predawn values as light and temperature increased (600–800 h) in all of the temperate species, with a 27, 16, 14 and 7.6% increase from predawn values in *Pinus*, *Pseudotsuga*, *Arbutus* and *Quercus*, respectively. Mean leaf temperature and photosynthetically active radiation (PAR) increased over the same time period by 1.2, 0.33, 0.35 and 2.7 °C and 57, 28, 119 and 36 μmol m^{-2} s^{-1} for *Pinus*, *Pseudotsuga*, *Arbutus* and *Quercus*, respectively ($P < 0.05$ for all comparisons of leaf temperature and PAR at 0600 versus 0800 h). *Pseudotsuga*, *Pinus* and *Quercus* all displayed decreased midday K_{leaf} (about 66, 59 and 30% reductions, respectively), relative to maximum values (Figure 3) which closely corresponded to minimum Ψ_L for the day. However, *Arbutus* showed little decrease in K_{leaf} (about 15% reduction), even though Ψ_L dropped to about −2.0 MPa

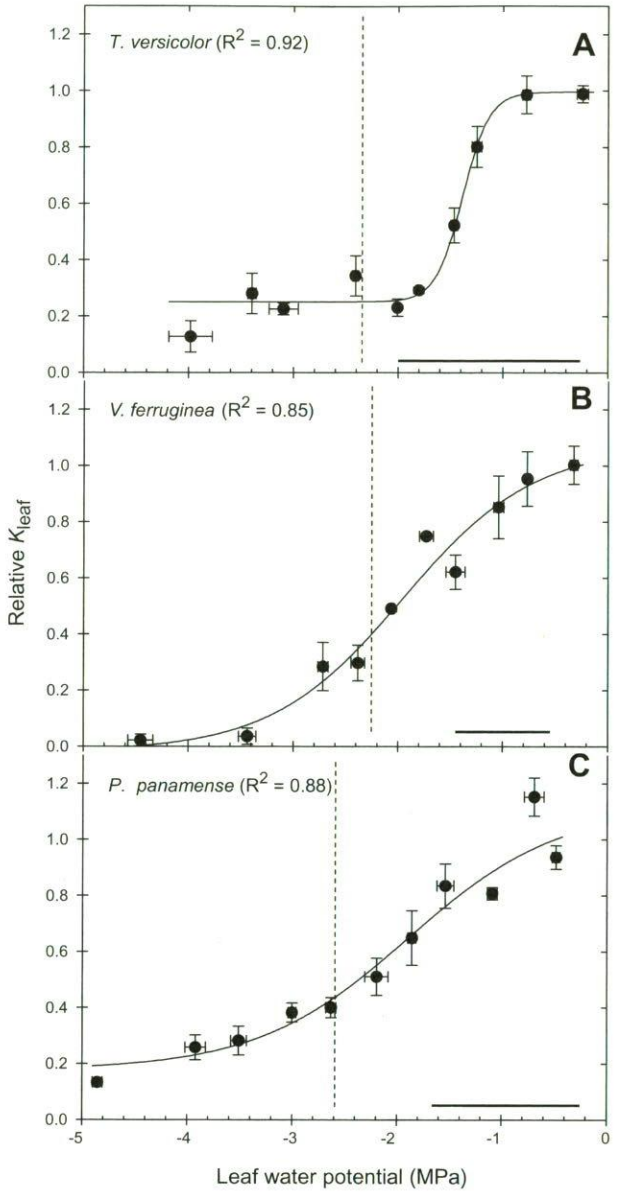

Figure 2. Leaf hydraulic conductance (K_{leaf}) measured in the laboratory for (A) *T. versicolor*, (B) *V. ferruginea* and (C) *P. panamense*. Dashed vertical lines represent mean bulk leaf TLP, horizontal bars represent observed ranges of water potentials in the field and vertical and horizontal error bars represent standard error.

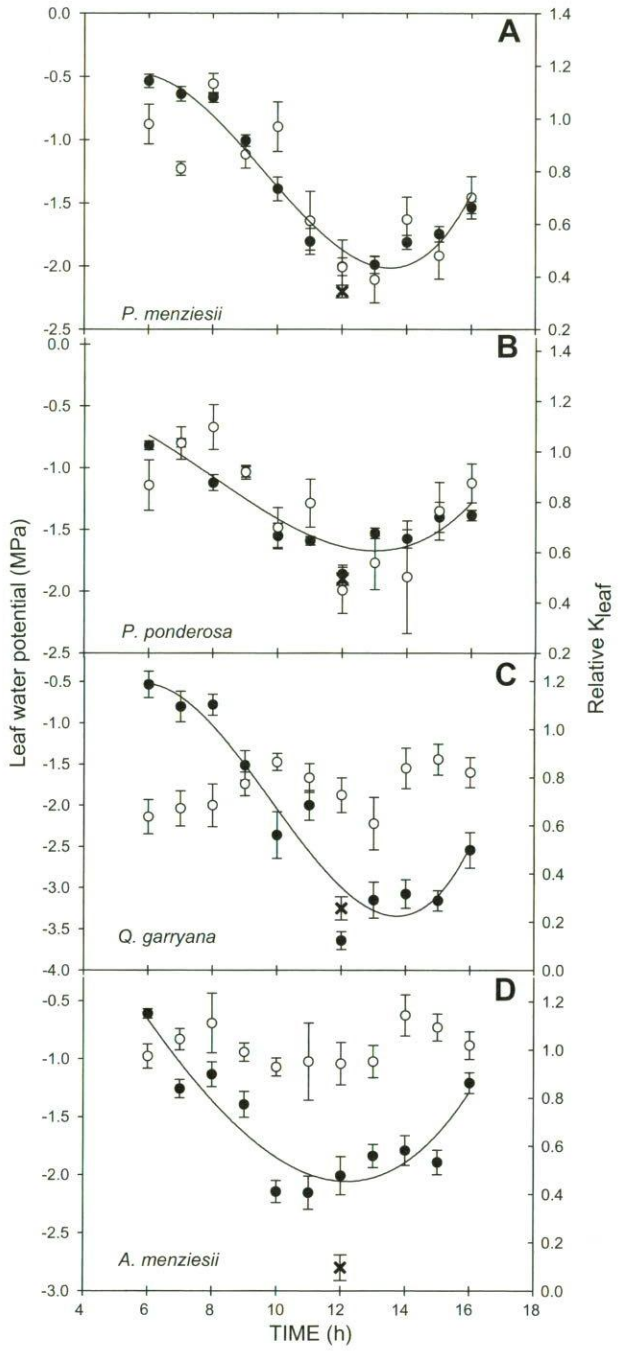

Figure 3. Leaf hydraulic conductance (K_{leaf}, open circles) and water potential (Ψ_{leaf}, closed circles) measured in the field for (A) *P. menziesii*, (B) *P. ponderosa*, (C) *Q. garryana* and (D) *A. menziesii*. Minimum Ψ_{leaf} (from Davis 2005), measured in the same area in 2002 and 2003, is represented by the '×' symbol. Curves are best-fit regressions fit through leaf water potential data using a cubic function ($y = y_0 + ax + bx^2 + cx^3$; R^2 values were 0.96, 0.85, 0.92 and 0.84 for panels A–D, respectively, and P values for all regressions were < 0.01) and vertical error bars represent standard error.

by midday. Although a large percentage of K_{leaf} was lost in both *Pseudotsuga* and *Pinus* by midday, there was substantial recovery by 1600 h. Midday relative K_{leaf} was 0.43 and 0.44 for *Pseudotsuga* and *Pinus*, respectively, but increased to 0.70 and 0.87, respectively, by 1600 h. *Quercus* K_{leaf} had completely recovered to morning values by late afternoon, although the midday K_{leaf} depression in *Quercus* (29%) was not as great as in *Pinus* or *Pseudotsuga*.

Mean minimum daily Ψ_L ranged from $-1.86(\pm 0.07)$ MPa for *Pinus* to $-3.64(\pm 0.11)$ MPa for *Quercus* and corresponded closely to earlier data collected in 2002 and 2003 by Davis (2005, see Figure 3). Large losses in K_{leaf} were predicted at midday from the laboratory-generated VC based on the minimum Ψ_L measured in the field for all

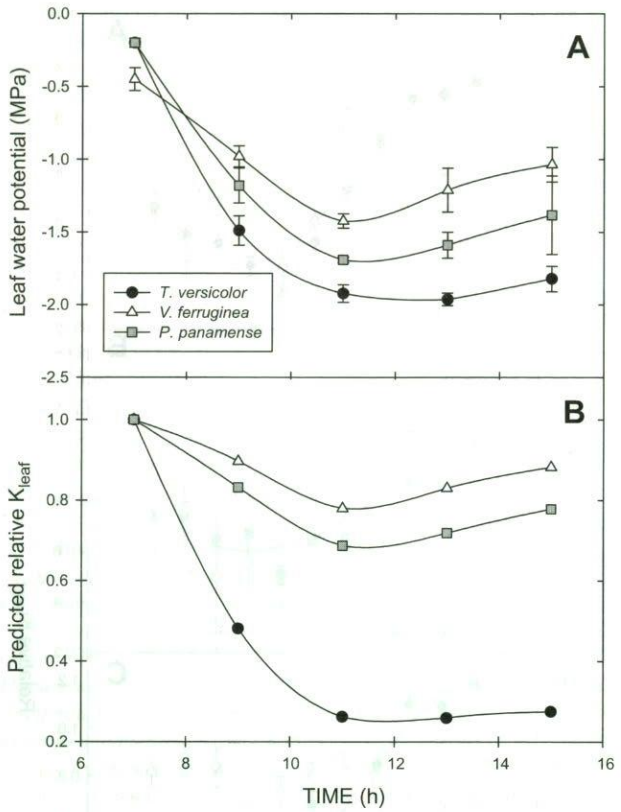

Figure 4. Measured field leaf water potential (Ψ_{leaf}, A) and predicted relative hydraulic conductance (K_{leaf}, B) for *T. versicolor*, *V. ferruginea* and *P. panamense*. Vertical error bars represent standard error.

species except *Arbutus* (Figures 1–3). In fact, in *Pinus*, *Pseudotsuga* and *Tachigalia*, mean minimum Ψ_L in the field corresponded to about the lowest point on the *y*-axis of the VC (the minimum K_{leaf}). Based on these water potential data (and leaf VC), K_{leaf} was predicted to decrease to about 78, 68 and 26% of maximum K_{leaf} by midday for *Vochysia*, *Protium* and *Tachigalia*, respectively (Figure 4). In addition, g_s in *Pseudotsuga*, *Pinus* and *Quercus* began to decline 2–4 h after K_{leaf} had already started decreasing (see Figures 3 and 5).

Discussion

The phenomenon of decreasing K_{leaf} with increasing water stress is well documented (e.g., Nardini et al. 2001, Bucci et al. 2003, Brodribb and Holbrook 2006, Woodruff et al. 2007). There are multiple potential mechanisms for dehydration-induced declines in K_{leaf}, including xylem cavitation, xylem implosion/deformation (Cochard et al. 2004), changes to downstream element conductivity (e.g., mesophyll) via changes in turgor (Brodribb and Holbrook 2003) and even biochemical-scale phenomena affecting the water permeability of cell membranes (Heidecker et al. 2003, Luu and Maurel 2005). However, recent work, using the independent methods of acoustic emission (Kikuta et al. 1997, Johnson et al. 2009) and cryo-SEM (Woodruff et al. 2007, Johnson et al. 2009), has shown that leaf xylem embolism is likely a critical element in the depressions in K_{leaf} associated with dehydration.

Few studies have characterized diel cycles of decline and recovery of K_{leaf} in situ. Bucci et al. (2003) found diurnal cycles of loss and recovery of hydraulic conductivity in petioles of *Schefflera macrocarpa* (C. & S) Seem. and *Caryocar brasiliense* Camb. In addition, Brodribb and Holbrook (2004) also observed decline and recovery of K_{leaf} over the course of a day and were able to predict loss

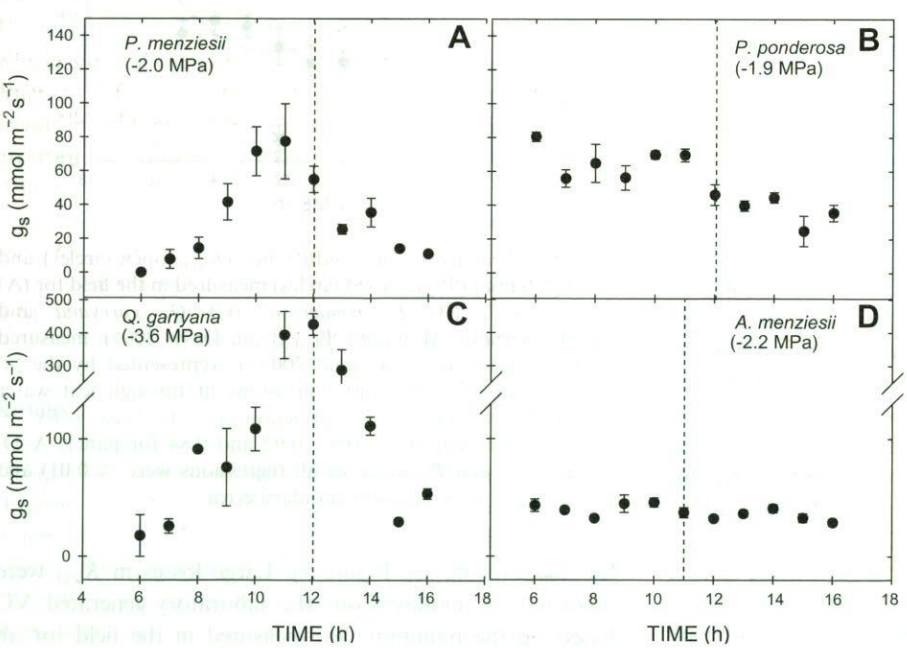

Figure 5. Stomatal conductance (g_s) measured in the field for (A) *P. menziesii*, (B) *P. ponderosa*, (C) *Q. garryana* and (D) *A. menziesii*. Vertical error bars represent standard error and dashed lines represent the time corresponding to daily minimum Ψ_{leaf}. Numbers in parentheses represent the actual minimum values of Ψ_{leaf}.

Table 2. TLP, pre- and post-TLP capacitance (C) and maximum leaf hydraulic conductance (K_{leaf}) in both the laboratory and field. TLP and C-values from pressure–volume analyses. Numbers in parentheses are standard errors (* and ** indicate significantly different field and laboratory maximum K_{leaf} at $P < 0.01$ and 0.001, respectively).

Species	TLP (MPa)	Pre-TLP C	Post-TLP C	Maximum K_{leaf}	
		mol m^{-2} MPa^{-1}		Field	Laboratory
				mmol m^{-2} s^{-1} MPa^{-1}	
Q. garryana	−3.63(0.24)	0.33(0.10)	1.74(0.15)	8.56(0.07)	6.00(0.07)**
A. menziesii	−2.40(0.20)	0.70(0.04)	2.73(0.15)	13.72(0.21)	13.35(0.10)*
P. menziesii	−2.30(0.21)	0.44(0.01)	0.98(0.02)	7.40(0.08)	3.94(0.18)**
P. ponderosa	−2.23(0.14)	0.55(0.02)	1.11(0.12)	8.15(0.17)	5.28(0.15)**
T. versicolor	−2.39(0.08)	0.56(0.09)	2.35(0.46)	n/a	25.56(0.74)
V. ferruginea	−2.23(0.08)	0.60(0.11)	2.52(0.30)	n/a	28.28(0.64)
P. panamense	−2.57(0.08)	1.62(0.29)	6.80(0.73)	n/a	13.45(0.56)

and recovery of K_{leaf} based on field leaf water potentials and laboratory-generated VC. The data presented here also show close agreement between laboratory and field K_{leaf} VC. Using laboratory-generated K_{leaf} VC and field leaf water potential measurements, it was predicted that *Tachigalia*, *Protium* and *Vochysia* all experience daily declines and recovery of K_{leaf}, although the degree of recovery in *Tachigalia* was small by the afternoon. Consistent with this, Meinzer et al. (2004) reported diurnal variation in K_{leaf} in the tropical species *Schefflera morototoni* (Aubl) and *Cordia alliodora* (Ruiz & Pav.) based on in situ measurements of branch transpiration and differences in Ψ_L of covered and exposed leaves. It should be noted, however, that the actual predicted percentage decrease and recovery of K_{leaf} in this study, based on laboratory-generated data, may be somewhat biased, due to potential differences in laboratory and field K_{leaf} (see Figure 1, panel A).

In this study, K_{leaf} began to recover from midday to late afternoon while leaves were still experiencing water potentials of less than −1 MPa. This phenomenon of restoration of conductance while the xylem is still under tension has been observed in other species as well. For example, Bucci et al. (2003) observed afternoon recovery of conductivity in petioles, and Nardini et al. (2008) found complete recovery of sunflower vein embolism at −0.33 MPa. There is much debate in the current literature concerning the mechanism(s) of recovery of conductance in plant organs experiencing negative xylem pressure (see Clearwater and Goldstein 2005, for review). Several hypotheses suggest that there may be mobilization of solutes to embolized conduits or regions adjacent to embolized conduits to induce refilling, and thus restore conductance (Grace 1993, Canny 1995, 1998, Hacke and Sperry 2003, Pickard 2003); however, more recently-proposed mechanisms implicate aquaporins or abscisic acid (ABA) or both as playing large roles in embolism recovery (Lovisolo and Schubert 2006, Lovisolo et al. 2008). Aquaporin expression is upregulated in many plants with increasing water stress (see Tyerman et al. 2002, for review) and it is thought that ABA may trigger expression of aquaporins during drought stress (Liu et al. 1994, Malz and Sauter 1999). It has been hypothesized that ABA may have dual roles in embolism recovery/repair: (1) by maintaining stomatal closure, thus preventing transpiration (Lovisolo et al. 2008) and (2) by triggering aquaporin expression in tissues adjacent to embolized conduits, which could induce radial water flow into the dysfunctional xylem (Kaldenhoff et al. 2008).

In the current study, there were large differences between maximum K_{leaf} measured in the field and in the laboratory, especially in *Pseudotsuga* and *Pinus* (88% and 54% increase in field K_{leaf} measurements as compared to those performed in the laboratory). Additionally, midmorning (800 h) K_{leaf} was about 16% greater than predawn K_{leaf} in *Pinus*, *Quercus* and *Arbutus* (and potentially *Pseudotsuga*, although K_{leaf} appeared to decrease in *Pseudotsuga* at 0700 h), even though leaf water potential was more negative than at predawn. Leaf temperatures increased over the same time period by $\sim$ 1.1 °C and PAR increased by $\sim$ 60 μmol m^{-2} s^{-1}. This trend in an early morning increase in conductivity has also been observed in petioles of two savanna tree species (Bucci et al. 2003), leaves of several temperate tree species (Lo Gullo et al. 2005) and leaves of sunflower (Tsuda and Tyree 2000). Leaf hydraulic conductance has previously been shown to increase with both increasing temperature and sunlight (e.g., Tyree et al. 2005, Scoffoni et al. 2008, Sellin et al. 2008, Voicu et al. 2008), which likely explains the observed early morning trends in K_{leaf} observed here, and potentially the differences in maximum K_{leaf} observed in the laboratory (equilibrated in the dark) and in the field (no equilibration). In addition, although absolute values of K_{leaf} differed between the field and laboratory in this study, their relative values were generally similar at the same water potential.

Maximum stomatal conductance (and photosynthesis) appears to be tightly coupled to maximum K_{leaf} across plant life forms (Sack et al. 2003, Brodribb et al. 2005, 2007, Sack and Holbrook 2006). However, stomatal behavior, as it relates to declining K_{leaf}, varies between species. In other words, different plants have different 'safety margins' or differences between water potential at stomatal closure and water potential at some degree of loss of K_{leaf} (see Sack

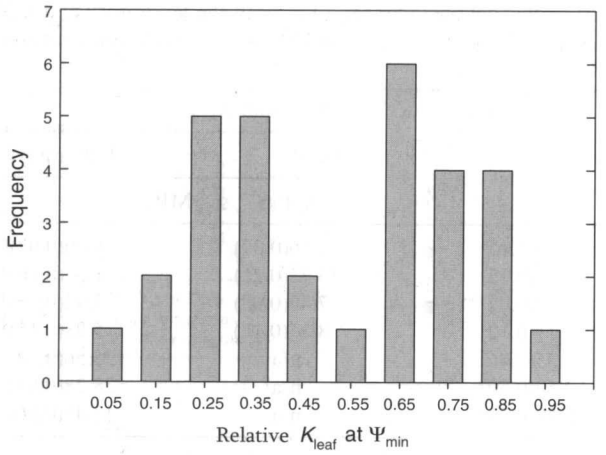

Figure 6. Frequency distribution of compiled data from this and other studies (see text for references). Relative leaf hydraulic conductance (either as measured, calculated from transpiration values and water potential differences between transpiring and non-transpiring leaves, or as predicted based on published VC and minimum leaf water potentials) at daily minimum leaf water potentials (n = 31). Data were placed into bins representing intervals of 10% relative K_{leaf} (e.g., 0.00–0.09, 0.10–0.19).

and Holbrook 2006). In the current study, the more conservative *Quercus* and *Arbutus* appeared to have much larger safety margins than did either of the conifers, both of which lost a substantial portion of K_{leaf} before stomatal closure. Other examples of plants with both wide and narrow safety margins can be found (e.g., Nardini and Salleo 2000, Cochard et al. 2002, Bucci et al. 2003, Brodribb and Holbrook 2004, Woodruff et al. 2007). However, a survey of the literature, along with data from the current study, suggests that plants tend to fall in one of two groups (i.e., are bimodally-distributed, Figure 6): plants that retain the majority of their K_{leaf} at their minimum daily Ψ_L and plants that have lost the majority of their K_{leaf} at their minimum daily Ψ_L. There was no apparent association between the mode of regulation of minimum K_{leaf} and the phylogeny, leaf phenology, functional group or habitat type of the 31 species represented in Figure 6.

Based on the data presented here, the rehydration kinetics method was a reliable method for making many estimates of in situ K_{leaf} fairly quick. Although no equilibration was used in this study, an earlier study (Scoffoni et al. 2008) suggests that long equilibration times are not necessary for accurate measurements using this method. In addition, little variation in Ψ_L (typically no more than 0.1–0.2 MPa) was observed between leaves on the same branch in this study. However, *A. rubra*, an understory species experiencing frequent sunflecks, showed higher variability of K_{leaf} and no consistent diurnal trends, and there were larger variations in Ψ_L between leaves on the same branch (DMJ, unpublished data). Therefore, this method may only be suitable for certain species or species growing in certain environments (e.g., open areas with little patchiness in sunlight).

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